

CMU Animal Care Committee Standard Operating Procedures (SOPs)

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Standard Operating Procedures (SOPs) provide a detailed description of procedures commonly used by animal users. SOPs offer investigators an alternative to writing detailed procedures in their animal use protocol. The SOPs that will be used must be specified in the protocol by the Principal Investigator. A new SOP that has not yet been approved by the CMU ACC must be defined with the protocol. Any deviation from an approved SOP must be clearly described and justified in the protocol. Approval of a protocol indicates approval of the deviation from the SOP for that project only.

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1. Wild Fish Handling

This SOP outlines general guidelines for the handling of wild fish. These guidelines are to help reduce both the stress and physical harm experienced by the specimens that are captured and released for educational purposes.

General Handling

1. The fish must be handled with care to prevent damage to their protective mucus coating, and to prevent stress and injury.
 - a. Handling should only be done when necessary and the handler must wet their hands with water from the capture site.
 - b. Do not remove the fish from water without necessity and return to their containment containing water of the specimen's origin as soon as possible.
 - c. Hold larger fish by supporting their bodies. Do not hold the fish by the tail or the operculum. Also, avoid compressing the abdomen.
 - d. Be aware of signs of stress or injury throughout handling. Signs can include grasping, lethargy, rapid gill movement or any abnormal behaviour.
2. All cautions should be taken to prevent injuries related to handling and to minimize stress.
3. If a fish becomes injured beyond the possibility of recovery or treatment than the fish should be euthanized by MS 222 (Tricaine mesylate).

General Considerations for Long Term Capture

1. The fish must be in water like that of their natural habitat. The following components and their levels must be taken into consideration: salinity, pH, nitrogen, oxygen, temperature, and the water must be dechlorinated.
 - a. Proper live support systems should be put in place to ensure the levels stay consistent.
2. Research must be done to ensure that the species of fish is getting the proper nutrients they need.
3. If the animal is a social species, then multiple organisms should be captured to reduce their stress, while making sure their aquarium is large enough for the number of organisms.
4. Disinfectants should be used to reduce the spread of diseases between fish and fish and people. Bleach solution, potassium permanganate, Virkon, or iodine-based disinfectants will be used as required and deemed appropriate.
5. Quarantine of new organisms

Resources

1. https://www.mcgill.ca/research/files/research/108-fish_aquatic_amphibian_reptile_analgesia_-_march_2022_v2.pdf
 - University SOP
2. <https://noto.ca/wp-content/uploads/SOP-Working-Near-Wildlife.pdf>
 - "Safe Handling of Live Fish"
3. <https://ccac.ca/en/training/modules/fish-stream.html>
 - "Fish Stream" training module from CCAC
4. <https://environment.fiu.edu/facilities-research-groups/marine-research-facilities-fleet/aquatic-animal-care/>
 - Gave detailed list of the different aspects to consider
5. UNBC's Standard Operating Procedure W-02 Catching and Handling Wild Fish

2. Occupational Health and Safety Program Relating to the Handling of Wild Animals

Purpose: To ensure the safety and health of the individuals responsible and involved in the handling of the wild animals in both research and teaching scenarios

Policy: To limit the unnecessary risk that individuals will face when handling wild animals. Precautions will include the assessment of allergies and other medical conditions that may put an individual at risk during research and teaching exercises.

General Guidelines

To ensure the safety of research and education participants understanding and knowledge of previous and present medical conditions is vital. Important information includes allergies, vaccinations, asthma, etc. A risk assessment should be conducted for each research or teaching activity which will be presented to participants. Each participant must read the risk

assessment and complete an informed consent form on the potential risks involved with field work. Once participants understand the potential risks, they can choose whether to participate in the work or not, and they do not need to provide a reason or disclose any personal health information. Participants are encouraged to discuss any concerns they have with a health care practitioner. Risks include, but are not limited to the exposure to:

- Aquatic species
- Infectious agents
- Solar radiation
- Chemical Carcinogens
- Reproductive Hazards/Teratogens
- Environmental allergies such as pollen, grass, mold, dust, etc.
- Chemical allergies
- Asthma triggers/instigators such as physical exertion
- Both natural and chemical skin irritants (such as plants and latex gloves)

To prevent the transmission of zoonotic and other bacterial infections individuals should wear personal protective equipment (PPE) and avoid specific actions when handling wild animals.

- Do not eat, drink, apply makeup or use any form of smoking device
- Wear eye and respiratory protection when appropriate
- Wear gloves when handling aquarium water, animals, animal tissues, body fluids and waste, and wash hands after contact.
- Cover abraded skin, cuts, scrapes, and sores and do not allow wound contact with the, fish contaminated materials or the water containing the fish.
- Clean and disinfect equipment that comes in contact with wild animals

There are a variety of other safety risks associated with working outdoors with wild animals. Caution should be taken in regards to environmental risks such as insect bites and stings, working around bodies of water, uneven terrain, extreme temperatures, sun exposure, Man-made concern include scalpel, needle, and chemical usage, such as disinfectants and pharmaceuticals. Precautions that should be taken include,

- Use of insect repellent
- Use of sunscreen
- Wearing long pants, long-sleeve shirts, and hats to reduce sun exposure
- Bring sufficient water for the number of people and duration of field work
- Wearing closed-toe shoes with sufficient tread
- Have a minimum of two people working at a time
- Have a minimum of one person with first aid training
- Bring a standard field research First Aid Kit, provided by CMU
- Do not direct sharp instruments towards others or self
- Keep sharp instruments in protective cases until in use
- Those handling chemicals must complete WHMIS training and know how to look up the chemicals within the CMU chemical archive
- Keep chemical bottles closed and wear gloves when handling

Resources

1. <https://researchintegrity.asu.edu/animals/forms>
2. <https://www.uidaho.edu/-/media/UIDaho-Responsive/Files/research/Faculty/research-assurances/IACUC/Health/zoonoses-resources/zoonoses-fish-including-aquarium-fish.pdf?la=en&hash=F10D22C5F1003D72E9F6DD73B9080F968B923D25#:~:text=The%20Ozoonotic%20diseases%20associated%20with,Salmonella%2C%20Klebsiella%20and%20Streptococcus%20iniae>
3. <https://pubs.usgs.gov/tm/15/c02/tm15c2.pdf>

3. Euthanasia Procedures and Waste and Carcass Disposal

This provides a general outline for recommended euthanasia practices of fish and suggestions for safe and appropriate carcass disposal.

Euthanasia: Humane approaches to euthanasia for the welfare of fish should be efficiently and effectively employed wherever indicated. Reasons for humane euthanasia include:

- 1) Illness or injury where animals do not have a reasonable prognosis for recovery, or where:
- 2) Treatment is not a reasonable or humane option
- 3) Research design requiring animals to be sacrificed for necropsy and further study
- 4) Any other condition causing unnecessary suffering and stress

Euthanasia should be carried out in a manner appropriate to the species, size, and number of fish to be euthanized.

Protocols for euthanasia should include:

- 1) Identified individuals who have been properly trained in the employed method of euthanasia
- 2) Clear intervention and endpoint criteria
- 3) Plans for appropriate disposal of carcasses and waste water

Acceptable methods are those that result in rapid and irreversible unconsciousness and prompt death. The DFOs Euthanasia of Finfish Training Template may be used as a guide to outline appropriate euthanasia techniques. Persons performing euthanasia should be competent in the method used including confirmation of death:

- Loss of movement
- Loss of reactivity to any stimulus
- Flaccidity
- Respiratory arrest and cessation of rhythmic opercular movement
- Loss of eye-roll – the movement of the eye when a fish is rocked side to side

The procedure must be repeated or another method employed if signs of life and recovery are noted. Fish must be confirmed dead with multiple confirmatory endpoints before disposing.

Disposal: Carcasses must be disposed of in accordance to municipal, provincial, and federal regulations, and biosecure storage must be used until carcasses are disposed of. The method of euthanasia may dictate the means of disposal available.

Where drugs such as TMS (Tricaine Mesylate) are used, proper PPE must be used and waste water must also be properly disposed of.

Means of disposal may include

- 1) Burial in designated locations in accordance with local regulations
- 2) Incineration
- 3) Composting in industrial composters for animal or biohazard waste

Resources:

National Farm Animal Care Council Code of Practice for Farmed Salmonids

<https://www.nfacc.ca/farmed-salmonids-code-of-practice#>

Canada Department of Fisheries and Oceans Animal-User Training Template

[3.0 Euthanasia of Finfish \(ccac.ca\)](https://www.ccac.ca/3.0-Euthanasia-of-Finfish)

CCAC Guidelines On the Care and Use of Fish in Research Teaching and Testing

4. Endpoints

Purpose: To develop guidelines for establishing endpoints for the purposes of minimizing or reducing experimental animals' pain and/or distress.

Policy: Ensure that animals are not subjected to any unnecessary pain or distress and that the experimental design offers them every practicable safeguard, whether in research, in teaching, or in testing procedures, as outlined in the CCAC Policy Statement: Ethics of Animal Investigation.

Responsibility: Principal Investigator, Consulting Veterinarian, CMU ACC

The term "Endpoint" is defined as "the point at which an experimental animal's pain and/or distress is terminated, minimized or reduced, by taking actions such as killing the animal humanely, terminating a painful procedure, or giving treatment to relieve pain and/or distress", as defined in the *CCAC Guidelines on: Choosing an Appropriate Endpoint in Experiments Using Animals for Research, Teaching and Testing*.

General Guidelines

In experiments involving animals, any actual or potential pain, distress, or discomfort should be minimized or alleviated by choosing the earliest endpoint that is compatible with the scientific objectives of the research. Selection of this endpoint by the investigator should involve consultation with the Consulting Veterinarian and is subject to approval by CMU ACC.

Endpoints are an objective record of any deviations from an animal's "normal" status, followed by a correlation of these changes with degrees of discomfort, pain and/or distress.

Examples of endpoints may vary between species, and may include (but are not limited to):

- Illness (measurable clinical signs)
 - Signs of pain and distress
 - Irregular behavioural responses to external stimuli
 - Change in eating/drinking habits
 - Compromised motion
 - Bleeding
 - Irregular breathing
- Severe damage to fins or gills of fish due to trapping

CMU ACC is responsible for establishing the structure to ensure that the earliest endpoints consistent with producing reliable data are considered, identified, and used. Application of these endpoints is the joint responsibility of the Principal Investigator and the Animal Facility staff. The following information shall be obtained **by the Principal Investigator** to ensure that an appropriate endpoint will be in place:

1. What are the scientific justifications for using the proposed endpoint?
2. What is the expected time course for the animals, from initial treatment to first signs of pain/distress, to the death of the animal, based on previous information with the specific model under study?
3. When are the effects to the animal expected to be the most severe?
4. If the course of the disease and expected signs of the adverse effects are unknown, could an initial (pilot) study, under close observation by the Principal Investigator and/or Animal Facility staff, answer these questions?
5. Has a checklist of observations, on which the endpoint will be based, been established?
6. Who will monitor the animals (identify all responsible) and keep records?
7. Has a clear chain for reporting observations been established?
8. What will be the frequency of animal observations: a) during the course of the study; and b) during critical times for the animals?
9. Do the investigators, animal care and technical staff have the training and expertise to monitor the animals adequately?
10. What provisions have been made to deal with any animals that show unexpectedly severe signs and symptoms?
11. For toxicological studies, have existing toxicological data been evaluated?
12. What basic endpoint responses will be prescribed? (see below)

Basic Endpoint Responses

Principal Investigators are responsible for identifying endpoints, using the categories below. They must also indicate how the endpoints will be quantified, and the type of intervention that will be used for each endpoint. These must be detailed for each study within the protocol application. In addition, objective values should be assigned to relevant animal conditions and responses to reaching an endpoint shall be well defined in the protocol.

Light Mediation: Prescribed intervention is applied in an attempt to address the area of concern, but the animal is not removed from the study group on a trial basis.

Moderate Mediation: The animal is temporarily removed from the study to halt the escalation of pain or advancement of illness and eventually eliminate the cause of concern.

Heavy Mediation: The animal is permanently removed from the study but remains under care with hopes to use the individual animal with a future purpose in mind provided concerns are fully remedied.

Euthanasia: The animal is euthanized with the understanding that the concern had progressed to a point beyond intervention, remedy, and to a level causing significant distress to the animal.

The CMU ACC may respond with recommendations and directions on how to improve endpoints set for conditional approval of a study. Personnel working with animals at all levels should always be aware of any actual or potential pain, distress, or discomfort and change in normal animal condition. When detected, any signs indicating pain or potential illness should be logged on the animal's individual care sheet and all staff should be made aware of the log entry and the contents therein. The Principal Investigator should be notified when an endpoint response is applied due to an endpoint being reached.

Field Research

Due to a lack of Animal Care personnel in contact with animals on a daily basis, the Principal Investigator of field work shall clearly define a chain of reporting observations and individuals responsible for mediation where necessary. The Principal Investigator shall be contacted, when not present, at such time that an endpoint is reached. It is the responsibility of the Principal Investigator to make the decision of when it is necessary to use heavy mediation or euthanasia. In this case, documentation detailing the rationale for using the particular response must be provided to the veterinarian within one week of the occurrence.

5. Crisis Management

Background

This Crisis Management Program is developed in conjunction with other CMU institutional crisis management plans. The term crisis is defined as any unplanned event which triggers a real, perceived or possible threat to the life, health or safety of animals and/or personnel, or to the institution's credibility.

CMU's Animal-related Crisis Management Team

1. VP Academic
2. Communications and Marketing Director

3. VP Finance and Administration
4. Laboratory Steward
5. Occupational Health and Safety Chair
6. Biology faculty member and Animal Care Committee (ACC) member

Crisis Management Procedures

- A. As outlined by the CCAC, the specific types of crises related to animal care and use programs can involve any of the following:
 - a. Fire, chemical spill or explosion
 - i. As CMU does not have any animal facilities on campus, no specific animal-related mitigation plans are required. CMU's Laboratory Safety Manual will be adhered to in the event of a fire, chemical spill, or explosion in the lab.
 - b. Natural disaster, power outage
 - i. In the case of minor disruptions, back up generators will provide power for basic lighting as well as all freezers in the biology laboratory.
 - c. Break-in, vandalism, unauthorized removal of animals
 - i. In the case of a break-in or vandalism, personnel will phone 911 if they have entered the premises and have noticed that this has occurred. Personnel will notify authorities as to the location and will not touch anything until emergency personnel have arrived. If there is still evidence of someone on the premises, CMU personnel will leave immediately and notify 911. If CMU personnel notice anyone in the process of removal of prepared specimens, they will call 911. Once personnel are in a safe place, they will call the VP Operations.
 - d. Bomb threat
 - i. Contact the local police immediately by phoning 911.
 - e. A sit-in or barricade action, a negative media event requiring a response
 - i. CMU's Crisis Communication Strategy will be followed in the event of an animal-based crisis.
 - ii. As per CCAC's recommendation, there should be no contact with the media/public until all relevant facts of the animal-related crisis are known.
 - iii. CMU's Communications and Marketing Director will be the principal spokesperson for media and public inquiries.

Field-work Hazards

In the event of a crisis involving a student or faculty member while engaged in off-site field work, CMU's *Crisis Communication Strategy* will be adhered to. If there is an emergency, CMU personnel will call 911 if appropriate.